BREEDING AND SELECTION PAPERS

Breeding orange-fleshed sweetpotato varieties for East Africa

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Abstract

The area under sweetpotato is expanding faster than any other major food crop in sub-Saharan Africa (SSA) and yet vitamin A deficiency (VAD) is a chronic threat on the continent. Orange-fleshed sweetpotato (OFSP) varieties with high levels of beta-carotene (vitamin A) in storage roots exist for combating the widespread VAD. Minerals and vitamins in food staples eaten globally by the poor may be increased by conventional plant breeding or by use of transgenic techniques, a process known as biofortification. One of the requirements for biofortification to be successful is that the breeding must be successful, which means that high nutrient density must be combined with high yields and high profitability. The International Potato Center (CIP) and collaborating partners introduced approximately 200 sweepotato genotypes from CIP, Lima, Peru, to East Africa over the last decade. CIP and the partners also conducted a genotype by environment (GxE) OFSP trial including mostly landraces between 2004 and 2006 to standardize sweetpotato breeding protocols by collaborators. Although some OFSP landraces had high dry matter (about 30%) desired by consumers and were released in different countries, they did not have high field sweetpotato virus resistance, very much required in the region. Promising OFSP clones from the field tests were included in crossing blocks at Namulonge in Uganda. The program at Namulonge produced breeding populations, a large proportion, about 40%, of which was sent to 14 collaborating SSA countries. Some of the clonal selections from these populations are about to be released by different countries. Currently, it takes 7-8 years to breed deep OFSP varieties adapted to local conditions because the high beta-carotene must be combined in the same genetic background with high yield and other desirable important traits. To shorten the breeding cycle, CIP proposes to centrally generate by population improvement diverse populations for national programs to use to produce a wide range of varieties that have the preferred trait combinations required by farmers and consumers in their countries. The proposed breeding scheme will use "accelerated breeding", exploit heterosis, and use molecular markers to achieve rapid progress in producing new adapted sweetpotato varieties.

Keywords: Sweetpotato virus disease, SPVD, *Alternaria bataticola* blight, biomass yield.

Introduction

The area under sweetpotato is expanding faster than any other major food crop in sub-Saharan Africa (SSA) and yet vitamin A deficiency (VAD) is a chronic threat on the continent. Orange-fleshed sweetpotato (OFSP) varieties with high levels of beta-carotene (provitamin A) in storage roots exist for combating the widespread VAD. Malnutrition among young children is increasing in SSA. Vitamin A is an essential micronutrient for humans. Globally, 127 million children under six years of age are estimated to be affected (West, 2002). SSA and India have the highest estimated prevalence rates of sub-clinical VAD. VAD limits growth, weakens immunity, causes xeropthalmia leading to blindness, and increases mortality (Sommer and West, 1996). There are two forms of vitamin A available in foods: preformed retinol (vitamin A) found in animal foods such as milk, liver, milk and eggs, and provitamin A carotenoids obtained from plant foods such as yellow and orange vegetables, dark green leafy vegetables, and fruits, and OFSP (McLaren and Frigg, 2001). β-carotene is the major provitamin A carotenoid, and the main carotenoid in OFSP. Poor households normally cannot afford to consume the highly bioavailable animal foods in their normal diet. High rates of deficiency in the major micronutrients (vitamin A, iron, and zinc) are prevalent among poor populations that depend on plant-based diets (Hess

et al., 2005). Most plant sources of vitamin A are seasonal, and after provitamin A carotenoids are absorbed into the body, they must be converted into retinol for use by the body. Rates of conversion vary among carotenoid containing plant foods (up to five fold) and also depend on other foods consumed at the same time (e.g., fat increases absorption) and the health status of the individual. More deficient individuals absorb and/or convert at higher rates than replete individuals. Heat processing may also enhance conversion rates compared to the raw product, depending on the plant matrix (Hess et al., 2005). Current guidelines express provitamin A activity in Retinol Activity Equivalents (RAE). In the RAE definition it is assumed that 16.7% of the ingested beta-carotene is absorbed and 50% is converted to retinol. The result is an average conversion factor of 12 units of betacarotene to form 1 RAE. Most dark green leafy vegetables are possibly less bioavailable than this, while palm oil is far superior (2:1 conversion factor). On the contrary, the conversion factor for preformed retinol from animal sources is 1:1 and for other provitamin A carotenoids 24:1 (Institute of Medicine, 2001). In a recent studies Africa, evidence was presented regarding the potential impact of OFSP on young child vitamin A status. The study demonstrated that OFSP is bioavailable and efficacious in improving vitamin A status in children (Jaarsveld et al, 2005). In another study, significant improvements in vitamin A intake and serum retinol concentrations (a proxy for vitamin A status) were obtained from an action-research project of an OFSP-based integrated agriculture-nutrition-market intervention in a very resource poor setting in Central Mozambique (Low et al., 2007). The study in Mozambique emphasized the importance of having the three important components (agriculture, nutrition and market interventions) to ensure improvement in young child vitamin A intakes and sustained adoption of the new material. In a third study, Haskell et al. (2004) using isotopic tracer deuterated retinol to estimate total vitamin A stores in 14 Bangladeshi men determined a conversion factor of 13:1 for OFSP when it was cooked pureed with a small amount of oil.

OFSP as a staple food has advantage over most vegetables because it can supply significant amounts of vitamin A and energy simultaneously, thereby helping to reduce both VAD and undernutrition. OFSP is an example of a biofortified crop in which the micronutrient status of staple foods is improved by plant breeding to the point where impact on micronutrient status can be achieved (Bouis, 2002). Because the poorest households normally obtained over 60% of their energy needs from food staples, this strategy is especially suited to poor rural households that cannot access purchased fortified food products but could grow OFSP. Due to the urgent need to address widespread VAD in SSA, the development and use of betacarotene-rich OFSP roots deserves special attention. One of the requirements for biofortification to be successful is that the breeding must be successful, which means that high nutrient density must be combined with high yields and high profitability.

Materials and Methods

Sources of OFSP: There were three sources of OFSP:

a) During 1990s, CIP began introducing OFSP clones collaborating with National Agricultural Research Institute (NARI) partners. Most existing varieties in SSA then were white or yellow-fleshed. SSA countries received approximately 200 clones as in vitro sweepotato plantlets directly or indirectly from CIP, Lima, Peru, via CIP, Muguga, Kenya (Mwanga and Ssemakula, 2010).

b) Twenty pathogen tested OFSP clones were sent in November 2002 as in vitro plantlets (2 per clone) from the International Potato Center (CIP), Lima, Peru to CIP, Muguga, Kenya, and Namulonge, Uganda. The 20 OFSP clones had been bred by CIP in Lima for high dry matter content. The clones were micropropagated in 2003 in tissue culture laboratories at Mugaga and Namulonge, and then propagated in aphid proof screenhouses. In 2004 an evaluation of the clones to establish adaptability was conducted in four locations in Uganda, at Namulonge, Kachwekano, Ngetta, and Serere (Table 1).

c) In 2004/2005 15 pathogen tested sweetpotato clones, most of them landraces and from different countries were sent by CIP Muguga, Kenya to different countries in SSA to conduct genotype by environment (GxE) trials. The clones were: Carrot C, (deep orange), Mayai (deep orange), and Ukerewe from Tanzania; 199062.1 (breeding line) from CIP, Lima; K135 (orange), Zambezi (deep orange) from Zambia; K566632, Pipi, K118, and Kakamega (SPK004) (yellow/orange) from Kenya, Ejumula (check for beta-carotene content), and NASPOT 1 (check for root yield) from Uganda, Resisto - originally from USA (check for beta-carotene content), Jonathan originally from USA (check for beta-carotene content). Based on

the results of trials in Uganda and GxE trials in 15 countries, the individual countries went to scale with promising clones, and countries with active sweetpotato breeding programs included the most promising clones in their crossing blocks.

In Uganda the the trials were planted in June/July 2004 following experimental details described below for the regional trial. Based on the performance of the clones in 2004 in Uganda, in 2005 mini vine cuttings of seven promising clones in Uganda were sent from CIP Muguga to collaborators in seven countries, namely, Democratic Republic of Congo, Ethiopia, Kenya, Madagascar, Rwanda, Tanzania, and Uganda. Each country selected at least three trial sites representing low, 0 to 1,000 meters above sea level (m.a.s.l.), mid, 1,000 to 1,500 m.a.s.l., and high, above 1,500 m.a.s.l. altitudes. The site selection was based on altitude or clearly distinct agro-climatic conditions.

OFSP GxE regional trials: In the GxE trials the clones were selected based on the availability of OFSP vines in each country. SPK004 was used as a common check because it had been already introduced in the participating countries before the conduct of the experiment. Mini cuttings were propagated in the field. Planting materials were vine tip cuttings, about 30 cm long. Each clone was planted in 4 rows, 1 m apart, 6 m long, 0.3 m between plants (33,300 plants ha^{-1}) in 3 replications in a randomized complete block. The trials were planted between mid to late 2005, and were harvested 4-6 months after planting depending on elevation. Standard data sheets were provided for collecting data during the growth period on establishment, vigour, sweetpotato virus disease (SPVD), Alternaria blight, and at harvest on stand count, vine weight, and root characteristics, including taste and acceptability. The middle rows of each plot were harvested for data analysis. For the GxE data were analyzed only for countries and sites that had complete data sets. Sites and countries that had missing clones or inconsistent data were excluded from the analysis. Information on status of crossing blocks, number of clones at different stages in the sweetpotato breeding cycle in each country was obtained from reports during an annual sweetpotato breeding meeting in Mukono, Uganda, in June 2010.

Results

Almost 100% of the OFSP clones received and evaluated in different agroecologies in Uganda were not adapted to the growing conditions. The clones were not suitable for local consumption; they had low dry matter content (DMC); were susceptible to *Alternaria bataticola* blight at high altitude, and at lower altitudes under high SPVD pressure, the most devastating disease of the crop in SSA (Table 1). Promising clones were included in crossing blocks at the National Crops Resources Research Institute (NaCRRI) at Namulonge and other countries in East Africa. Promising OFSP clones (Table 2) were released and disseminated in different countries (Table 3). The Uganda National Sweetpotato Program at NaCRRI under the National Agricultural Research Organization (NARO) in collaboration with the PRAPACE Network, was responsible for improving sweetpotato for SPVD resistance. The program focused on combining desirable traits in a genetic background with SPVD resistance; it generated breeding populations (seed) between 2002 and 2009. About 20-40% of the seed was sent to collaborating countries, Burundi, Ethiopia, Rwanda, Kenya, Tanzania, Madagascar, Ghana, Nigeria, Malawi, Mozambique, Zambia, South Africa, and Burkina Faso. For example, about 471,400 seed was sent out from Uganda in 2007/2008 to 14 collaborating countries in SSA. The parents in the crossing blocks were exploited to produce populations that combine important traits such as resistance to SPVD and *A. bataticola* blight, high -carotene concentration; high DMC (30% or more); good root shape; and high biomass. Table 3 shows the current status of OFSP clones in East and Central Africa as a result of screening, evaluation, and selection from introductions, landraces, and breeding materials. Varieties Kakamega, Ejumula, NASPOT 9 O (Namulonge sweetpotato orange-fleshed) and NASPOT 10 O have been introduced into several African countries.

Discussion

Although some OFSP landraces had high dry matter (about 30%)(Table 2) desired by consumers and farmers and were released in different countries (Table 3), they did not have high field sweetpotato virus resistance, very much required in the region. Promising OFSP clones based on field evaluations were included in crossing blocks at Namulonge in Uganda and other African countries. The program at Namulonge produced breeding populations, a large proportion, about 40%, of which was sent to 14 collaborating SSA countries. Some of the clonal selections from these populations are about

to be released by different countries. Currently, in countries with two seasons in a year, it takes 7-8 years to breed deep OFSP varieties adapted to local conditions because the high beta-carotene must be combined in the same genetic background with high yield and other desirable important traits (Andrade et al. 2009; Grüneberg et al. 2009). To shorten the breeding cycle, CIP proposes to centrally generate by population improvement diverse populations for national programs to use to produce a wide range of genotypes that have the preferred trait combinations required by farmers and consumers in their countries. The proposed breeding scheme will use "accelerated breeding" to reduce the sweetpotato breeding cycle from eight to about four years (Grüneberg et al. 2009). The accelerated sweetpotato breeding method which involves rapid multiplication of genotypes from seedlings, and early evaluation of the genotypes and families in multiple environments, is also used by national programs for selecting new varieties. Near infrared spectroscopy (NIRS) will be used for rapid analysis of quality attributes (Andrade et al. 2009). In population improvement, heterosis will be exploited, and development, validation and use of molecular markers will complement the efforts to achieve rapid progress in producing new adapted sweetpotato varieties.

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Table 1. Performance of 2nd batch CIP orange-fleshed sweetpotato clones at Namulonge, Kachwekano, Ngetta, and Serere, October 2003 March 2004.

Serial No.	CIP code	Clone name or pedigree	Site				Mean across sites	SPVD[1] at Namulonge	Flesh color
			Namulonge	Kachwekano	Ngetta	Serere			
			Storage root yield (t/ha)						
1	199004.2	CC89.147. 4 xOP	21.5	41.4	32.1	33.5	38.5	4.5	PO
2	199004.3	CC89.147. 4 x OP	25.0	26.2	26.9	11.5	29.1	4.3	DO
3	199005.1	CHGU 1.002 x OP	24.5	32.2	14.3	0.3	26.2	3.5	PY
4	199015.1	LM92.032 x OP	34.1	31.6	33.8	8.6	27.0	4.3	PY
5	199015.1	LM92.032 x OP	31.2	38.3	46.1	3.3	29.7	4.8	DO
6	199024.1	SR91.109 x OP	32.0	17.9	24.1	40.8	28.7	4.3	DO
7	199024.2	SR91.109 x OP	35.9	27.5	29.7	9.7	25.7	4.3	PO
8	199025.2	SR92.095. 3 x OP	25.0	41.1	57.1	22.2	42.6	4.0	DO
9	199026.1	SR92.095. 8 x OP	33.0	33.6	57.1	21.6	44.7	4.3	Y/O
10	199027.3	SR92.095. 10 x OP	11.2	46.6	49.5	15.7	30.8	4.0	Y/O
11	199034.1	SR95.628 x OP	28.3	27.5	21.4	27.8	26.2	5.0	PY
12	199035.1	SR95.636 x OP	24.5	32.1	22.8	20.6	32.1	4.3	DO
13	199035.5	SR95.636 x OP	36.8	26.6	47.6	5.6	41.6	5.0	DC
14	199035.7	SR95.636 x OP	20.0	41.7	57.1	5.9	37.9	4.0	PO
15	199035.8	SR95.636 x OP	37.8	39.1	25.0	14.6	29.1	5.0	PY
16	199057.4	LM94.422 x OP	35.0	50.0	16.4	11.1	28.1	4.3	PY
17	199062.1	SPV 78.001.3 x OP	35.0	30.4	40.7	5.6	37.1	3.3	PO
18	440203	Unknown	48.2	20.0	27.2	26.0	30.3	3.8	C
19	440443	Nashu 88 (81-88)	40.0	25.0	31.0	10.8	36.7	4.3	Y/O
20	420020	Huarmeyano	23.6	13.8	13.8	10.0	15.3	3.5	DY
21		Tanzania (LC[2])	41.3	23.1	16.8	19.6	26.4	2.8	PY
22		Dimbuka (LC)	40.1	24.4	21.8	50.0	47.2	3.5	C
23		New Kawogo (LC)	38.0	NA	NA	NA	NA	2.0	C
24		Kyabafuruki (LC)	49.3	NA	41.7	21.6	NA	3.8	W
25		Magabali (LC)	NA[3]	52.4	NA	NA	NA	NA	W
26		Otada (LC)	NA	NA	10.9	NA	NA	NA	C
27		Araka (LC)	NA	NA	NA	33.3	NA	NA	W
Mean			32.1	32.3	31.9	17.9	32.3	4.0	NA
LSD(0.05)			10.7	15.1	9.2	9.6	7.7	0.8	NA
CV (%)			23.7	33.1	20.6	38.1	34.3	15.7	NA

[1]SPVD (sweetpotato virus disease) rating scale = 1-5: 1 = no apparent damage/not present, 2 = very little damage/few present, 3 = moderate damage/numbers present, 4 = considerable damage/numbers, 5 = severe damage/very high numbers. Flesh color: W = white, C = cream, PY =pale yellow, DY = dark yellow, Y/O = yellow with orange or vice versa, PO = pale orange, DO = dark orange. NA = not applicable

Table 2. GxE root yield (t/ha) and dry matter (DM%) of orange-fleshed clones, 15 locations, 2006/2007

Clone		Ango oni	Gu rue	Umbe lezi	Zanzi bar	Buko ba	Kaka mega	Nge tta	Namu longe	Sere re	Rubo na	Kibu ngo	Kiba ha	Hom bolo	Man doto	Mim osa	Clone mean	Flesh color	DM
1	Carrot-C	2.8	12.0	13.2	1.1	4.1	23.3	6.4	32.2	27.9	2.5	3.6	3.5	5.5	12.2	3.5	10.3	O	29.8
2	K135	1.5	6.3	5.5	1.6	3.4	17.1	7.6	32.3	17.0	4.3	5.2	2.3	12.6	5.4	5.8	8.5	Y/O	30.9
3	Gweri	3.0	10.2	2.6	2.6	3.0	17.9	4.4	20.4	15.2	2.4	2.7	4.8	4.5	1.8	5.1	6.7	Y/O	33.0
4	Zambezi	2.8	15.7	14.0	2.8	8.1	15.4	1.7	22.9	5.8	3.2	4.1	3.6	4.3	15.7	4.7	8.3	O	33.0
5	Ukerewe	3.0	13.5	14.1	0.7	7.3	26.3	0.5	10.7	10.6	3.5	4.0	5.2	2.8	13.3	4.7	8.0	Y/O	34.0
6	Mayai	2.2	10.2	17.3	0.6	4.9	26.3	3.4	15.6	41.6	2.1	3.2	5.4	0.9	11.3	5.0	10.0	O	29.4
7	K566632	2.0	10.1	10.0	3.5	7.4	28.9	3.9	23.7	47.9	2.1	3.0	4.5	6.6	12.5	4.4	11.4	DO	32.7
8	K118	1.8	17.4	6.4	1.3	4.1	20.5	18.1	36.7	40.0	2.9	2.7	2.6	6.5	10.2	5.9	11.8	O/Y	29.3
9	Ejumula	2.8	19.4	13.6	6.4	5.6	33.1	2.0	12.6	11.0	2.1	2.2	2.6	4.0	16.1	4.3	9.2	O	33.3
10	Pipi	2.0	15.2	10.6	1.4	6.1	20.0	11.5	33.1	20.8	2.3	3.0	4.8	6.7	13.1	6.8	10.5	C	27.5
11	SPK004	0.7	12.1	10.1	1.0	5.0	17.3	11.5	28.1	26.8	3.2	4.1	5.0	2.5	8.9	4.3	9.4	O/Y	30.7
12	199062	3.0	16.0	15.5	1.3	9.7	26.7	2.8	28.1	24.1	3.7	4.7	7.1	3.8	20.3	3.1	11.3	Y/O	30.3
Site mean		2.3	13.2	11.1	2.0	5.7	22.7	6.2	24.7	24.1	2.9	3.5	4.3	5.1	11.7	4.8	9.6	NA	31.2
LSD(0.05)		1.3	8.1	9.0	1.8	2.4	10.2	5.2	13.0	12.7	1.8	1.9	3.5	6.7	3.2	0.9	1.9	NA	NA
CV		39.3	36.4	49.2	56.9	27.4	26.6	49.7	31.1	32.4	40.3	36	47.2	78.7	16.5	11.8	42.3	NA	NA

O = orange, O/Y = yellow with orange, C = cream

Table 3. Orange-fleshed sweetpotato in East and Central Africa (Eth= Ethiopia, Rw=Rwanda, Ke=Kenya, TZ=Tanzania, Ug=Uganda; DM= dry matter, O=orange, Y=yellow, DO=dark orange, SPVD=sweetpotato virus disease, Alt= Alternaria, S= susceptible, M=moderate, R=resistant, NA=not applicable)

Landrace	Country	Yield (t/ha)		DM	Root	B-carotene content/ 100g fresh weight		Resistance
		Root	Foliage	(%)	flesh color	mg	Retinol activity equivalent (RE)	to SPVD
Kakamega	Ug, Ke, Rw, TZ , Eth	14.9	42.0	32.1	O/Y	5.5	457.5	M
Ejumula	Ug, Ke, Rw, TZ , Eth	18.0	30.0	34.2	O	12.4	1032.5	S
Carrot C	Ke, TZ, Ug	10.4	NA	33.2	O	12.4	1032.5	S
K566632	Ke	11.7	25.5	21.3	PO	NA	NA	S
Mayai	Ke, TZ, Ug	17.0	12.7	33.0	O	12.4	1032.5	S
K135	Ke	11.7	33.4	27.7	Y/O	NA	NA	S
Gentute	Eth	17.8	35.4	27.8	DO	NA	NA	S
Kulfo	Eth	29.8	27.0	29.8	O	NA	NA	S
Damato	Eth	30.7	30.7	18.3	DO	NA	NA	S
Bred cultivars								
NASPOT 9 O	Ug,, Ke, TZ	19.9	22.6	30.1	O	11.0	919.2	R
NASPOT 10O	Ug,, Ke, TZ	19.8	25.7	30.5	O	11.0	919.2	R
SPK2001/261	TZ	13.2	8.3	31.0	O	NA	NA	M
SP KBH 03/069	TZ	8.8	6.3	35.0	O	NA	NA	M
Caceaperdo	Rw, DR. Congo	18.2	NA	29.5	DO	NA	NA	M
97/062	Rw	14.4	NA	28.4	O	NA	NA	M
2004/024	Rw	17.2	NA	30.8		NA	NA	M
Introductions								
Resisto (440001)	Ug, Ke, TZ	15.8	NA	27.0	DO	11.0	919.2	M
Tainung 65 (440189)	TZ	15.0	40.0	25.0	O	3.8	313.3	M
Zapallo (420027)	Ke, TZ, Eth	19.3	18.2	26.0	DO	NA	NA	S
199062.1	Ke, Rw, Ug, TZ, Eth	20.9	22.2	28.1	Y/O	NA	NA	M
Salybolo	Ke, TZ	11.0	15.2	27.5	O	NA	NA	S, Alt